# **National Aeronautics and Space Administration**

# PLANETARY PROTECTION ADVISORY COMMITTEE

May 29–31, 2003 Hilton Cocoa Beach Cocoa Beach, Florida

# **MEETING REPORT**

John D. Rummel	Norine E. Noonan	
Executive Secretary	Chair	

# PLANETARY PROTECTION ADVISORY COMMITTEE (PPAC)

Hilton Cocoa Beach Cocoa Beach, Florida May 29–31, 2003

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#### PLANETARY PROTECTION ADVISORY COMMITTEE (PPAC)

Hilton Cocoa Beach, Cocoa Beach, Florida May 29–31, 2003

Thursday, May 29, 2003

#### Welcome and Meeting Overview

Dr. Norine Noonan, Chair of the PPAC, called the meeting to order and welcomed the committee members, liaisons from other federal agencies, representatives from international space agencies, and other meeting attendees. She noted that the meeting would include program status updates and presentations on topics on which PPAC members had previously asked to be briefed. The launch of the first Mars Exploration Rover (MER-A) had been scheduled for May 31 and was the reason for holding the meeting in Cocoa Beach, near the launch site at the Cape Canaveral Air Force Station (CCAFS). However, the launch had been postponed until June 8. The PPAC would tour the Kennedy Space Center (KSC) on Friday afternoon (May 31) to visit the MER launch sites, as well as other sites of interest at KSC.

#### Report from the NASA Advisory Council

Dr. Noonan summarized the March meeting of the NASA Advisory Council (NAC), held in New Orleans. That meeting was taken up primarily with briefings and discussions about the Columbia Shuttle flight and the investigation into the Shuttle's loss during re-entry. Although the discussion had been lively, little of it was relevant to the work of the PPAC. Dr. Noonan described the NAC site visit to the NASA Stennis Space Center to observe a test firing of a Shuttle main engine. She noted that she would be able to present issues agreed upon during this PPAC meeting at the next NAC meeting, which starts on June 10, 2003. PPAC discussions on Friday, May 30, and Saturday, May 31, would be used to identify and formulate any issues or recommendations for this purpose.

Dr. Noonan welcomed new PPAC member Susanna Hornig Priest of Texas A&M University. The members then introduced themselves, noting their technical backgrounds briefly. Dr. John Rummel, Executive Secretary of the PPAC, described the routes by which PPAC advice reaches NASA managers and executives. The committee staff reviewed the logistics for the meeting, and Dr. Rummel noted several late changes to the agenda.

## Planetary Protection Program Status/Plans

Dr. Rummel said that the PPAC has been rechartered as an advisory committee to the NASA Administrator under the *Federal Advisory Committee Act* (FACA). Under the FACA rules, PPAC members will have 3-year terms, but terms cannot be renewed. This restriction does not apply to the Agency Representatives or International Representatives. Dr. Rummel then reviewed the NASA Planetary Protection Policy (NPD 8020.7E), which addresses both forward and back contamination. He presented a list of questions compiled by Dr. Debra Leonard after the October 2002 meeting and forwarded to Dr. Noonan (Appendix E). Dr. Rummel noted which questions would be at least partially addressed by specific presentations at this meeting and which would be addressed at future meetings.

Dr. Rummel then summarized the planetary protection (PP) issues currently before the PPAC, including issues on forward contamination (decontamination requirements for missions to different martian or outer planet environments), back contamination (acquisition, containment, and handling of samples returned to Earth), and implementation (new methods to monitor and remove microbial contamination). The issue of decontamination requirements for the outer planets has increased currency since the President's Budget Request for FY 2004, which includes funding for the Jupiter Icy Moons Orbiter (JIMO) mission. Mars remains an important issue for PP because Mars missions continue to make discoveries that surprise us and change assumptions about the martian environment. Dr. Rummel reviewed some of the recent evidence for water movement (erosion) on the surface of Mars and for the presence of water ice not only at the poles but at lower latitudes as well. The natural processes that have resulted over time in interchange of materials between Mars and Earth may have provided means for biological contamination already, in either direction. In this context, missions with PP implications include the Mars Express/Beagle 2 mission by the European Space Agency (ESA), to be launched in June 2003; the Mars Exploration Rover (MER) missions; Mars Reconnaissance Orbiter (MRO); the competed Scout missions, which may include a Mars orbiter or lander;

and the 2009 Mars Science Laboratory (MSL), which will have long-term heat sources on board its rovers. With respect to back contamination and sample handling, the draft Mars Sample Handling Protocol (discussed at the October 2002 meeting), is available on the NASA PP website. Dr. Rummel has been seeking approval from the NASA Office of General Counsel since November for publication of a *Federal Register* notice of the draft protocol. Dr. Noonan suggested that the PPAC consider, as a discussion topic on Friday, how best to transmit its concerns to NASA over the delay in formal public notice of the draft protocol.

With respect to control of forward contamination of Mars, better understanding is needed of microbial biodiversity of cleanroom environments, survival of Earth organisms under the range of conditions now anticipated on Mars, and a systematic approach to identifying and tracking the locations on Mars that may be capable of supporting Earth life. In implementation, the issues are developing and qualifying new methods to monitor microbial contamination on Mars spacecraft and to remove biological contamination from spacecraft. Dr. Rummel listed the 1992 recommendations from the Space Studies Board (SSB) of the National Research Council (NRC) on control of forward contamination of Mars and compared the standard NASA bioburden assay method (using culturation) with alternatives using enzyme-based assays or spore signatures. The PPAC discussed the Viking sterilization facility, which Dr. Rummel recently visited at KSC, and issues related to dry heat sterilization as the only currently approved method for the highest standard of spacecraft decontamination. The Genesis mission to collect solar wind particle and the Stardust mission to collect comet dust will be the next missions for which sample handling will be an issue.

During the subsequent discussion, Dr. Ronald Atlas asked about the level of oversight and independent verification NASA maintains with respect to PP on missions that are largely planned, developed, and conducted by contractors. Dr. Rummel described the oversight and review process that each mission project follows in submitting a mission plan with PP details, followed by multiple points during implementation when actual performance is assessed against that plan by NASA. NASA oversight includes assaying for contamination levels by non-project (independent contractor) staff. The assay results are incorporated into the prelaunch data required for flight readiness authorization. Bioburden issues (monitoring and decontamination) are addressed from the beginning of the process. Dry heat sterilization is in fact used for major portions of the equipment that cannot be opened and sampled. Each piece of a craft that will land on Mars, as in the case of the MERs, has a bioburden limit. However, problems can arise when materials are substituted that lead to unexpected changes in spore count. Overall, NASA does not assume that past successful performance by a contractor in meeting PP requirements means that oversight can be relaxed. Dr. Rummel described the working policy as "trust but verify," using independent validation/verification of what the mission project office and the project contractors report.

#### Mars Express/Rosetta Status

Dr. Gerhard Schwehm, the International Representative from ESA, updated the PPAC on the Mars Express and Rosetta missions. The Rosetta launch schedule was changed after the failure of an Ariane 5 ECA launch in December 2002. Dr. Schwehm discussed the issues that led to the decision to change the target from comet Wirtanen to comet Churyumov-Gerasimenko. A major remaining issue is that the Rosetta lander was designed for a smaller comet. The lander may not withstand the higher impact velocity from the stronger gravitational attraction during landing on the new target. The Rosetta spacecraft is still in a cleanroom. The mission delay has increased the budget by \$70 million, but ESA has agreements from the funding nations to rearrange budget elements to support a launch in February 2004. If launched then, Rosetta will reach Churyumov-Gerasimenko in 2014. At the next PPAC meeting, Dr. Schwehm will summarize results from cleanliness testing under the Ariane 5 shroud.

Mars Express is scheduled to launch on June 2. If the launch is delayed more than a day, the launcher will have to be refueled. The launch window extends to June 21. On the current schedule, Mars Express will reach Mars on December 25. 2003. Beagle 2, which will be ejected 5 days later, will land in Isidis Planitia, deploy solar arrays, and begin taking observations. The Mars Express orbiter will provide global high-resolution photogeology with super-resolution imaging of selected areas, global mineralogical mapping at 100 m resolution, global atmospheric circulation and composition, subsurface structure down to permafrost, surface–atmosphere interactions, and interactions of the upper atmosphere with the solar wind. The Beagle 2 lander will analyze the geology and mineralogy of the landing site, including organic and mineral

geochemistry. It will also search for life signatures (indirect approaches studying water presence, direct approaches using gas analysis, carbon isotope ratios, iron phases, and microfossils in minerals) and provide information on meteorology and climatology. In closing, Dr. Schwehmreturned to the issues of PP responsibility when different entities are involved, as in this mission, which the PPAC had discussed during the October 2002 meeting. The Beagle 2 team has had responsibility for PP preparation of the lander, with oversight by The Royal Society of London. ESA received the prelaunch PP report two weeks ago, and the ESA Director-General should sign it this week. ESA sees it as an ESA responsibility to submit the PP report to the international Committee on Space Research (COSPAR), but nondisclosure issues remain to be resolved with the Beagle 2 builders.

Dr. Schwehm next discussed the new ESA program for a 2009 launch of a large exobiology rover to Mars. Funding is not yet committed for the program, but ESA is asking the community to submit proposals. A small PP committee has been formed for the program, and PP will be developed in parallel with the mission's science program. The sponsoring program within ESA is Manned Space Flight, and it is an optional, rather than mandatory, program for the member states. If ESA can achieve an 80 percent budget subscription from member states, program scheduling will proceed. Dr. Schwehm suggested that PP activities coming out of this program would be of interest to the PPAC. For example, all the ESA member states still have separate national laws on environmental protection, and there is no legal framework for ESA, as a multinational entity, to state PP requirements for this type of mission. He believes, however, that the approval this week for a European Global Positioning System will bring the ESA closer to the European Union.

Dr. John Kerridge noted that if Beagle 2 were a NASA lander, it would have to be sterilized to the Viking standard (because it is a life-detection mission). In response to his question about who has ultimate responsibility for Beagle 2 with respect to PP, Dr. Schwehm said that, in ESA's view, ESA is responsible. The members then discussed whether Beagle-2 was a life-detection mission in the sense that would require heat sterilization (or an equivalent level of decontamination). Dr. Noonan remarked on the difficulties of achieving consensus in an international environment when no single entity, such as NASA, has control. Dr. Schwehm agreed that the situation would be different if Mars Express were a joint mission with NASA. A member of the audience commented that the Beagle 2 builders had consulted with U.S. PP experts and went beyond COSPAR Category IVa in their decontamination procedures.

#### Mars Exploration Program Status

Mr. Orlando Figueroa, Director of the Mars Exploration Program Office, reviewed the approved missions through FY 2009 and the planning for the Next Decade (FY 2010–2020) of Mars Exploration. The Mars Global Surveyor (MGS) remains in orbit, with 140,000 images returned so far from the Mars Orbiter Camera. Planning is underway for a third mission extension, to run through September 2008 (the current extension ends September 26, 2004). Mr. Figueroa emphasized the value of MGS for subsequent missions such as mapping the primary landing sites for MER at 2–6 m resolution. The Odyssey spacecraft is delivering large amounts of data (200 GB in January). In fact, the data rate is raising issues about the archive's ability to handle the data from the MRO mission. The prime mission for Odyssey ends in August 2004, and the Odyssey mission team is now preparing for the spacecraft's UHF relay phase in support of MER and Beagle 2. The use of Odyssey for data relay will allow MER to return twice as much data.

MER-A is on the launch pad and has been integrated with the rocket stage. The May 30 launch was delayed by discovery of an open fuse in the grounding circuit. A decision was made that the spacecraft can fly with the fuse open, but some questions remain about whether any other components were overstressed by the action that caused the fuse to open. If the current launch opportunity is missed, launch would be delayed by more than a year. Mr. Figueroa and the PPAC discussed the selected target sites for the two rovers and the rationale for site selection. The highest priority site is Meridiani, just south of the martian equator. Mr. Figueroa reviewed the technical capabilities of the rovers and their instrument suite, such as the PanCam (which provides the equivalent of 20/20 vision from 5 ft. above the surface), the contact instruments for spectrometry, and other imaging instrumentation. Each MER rover weighs 4–5 time more than the small Sojourner rover.

The MRO mission in 2005 begins the second cycle of seeking/in situ explorations. Mr. Figueroa described the role of NASA's Planetary Protection Office in reviewing PP plans and tests conducted by the project team. He addressed a question on the mission value of MRO versus Mars Express in terms of ability to do more mineralogy from orbit. MRO will have nine instruments, with a high-resolution imager and spectrometer as its two major instruments. For the Scout mission to be selected for 2007, the seven contenders include a lander, flyers, and a sample return mission. Due to resource constraints, NASA will select only 1 mission.

The MSL (Mars Science Laboratory), scheduled to launch in October 2009, will include remote sensing instruments and contact instruments like those on the MERs, but with better capability, in addition to analytical instruments not on the MERs. The landing site will be within the  $\pm 60^{\circ}$  latitude band. The nuclear power plant that enables its extended roving (several kilometers) and science research capabilities will also provide a mission life longer than 500 sols. The Phase A Concept Review is in August 2003, with a go/no go decision on Phase B in early 2005. The Announcement of Opportunity (AO) for the MSL instrument suite will be released in March 2004. Prototypes for the sample acquisition and handling devices are being constructed now at Jet Propulsion Laboratories (JPL). In response to a question from Dr. Klein, Mr. Figueroa and the PPAC discussed the constraints on introducing new science capability after the initial instrument selection. In effect, the selected developers can subsequently incorporate new technology in an instrument, as long as the interfaces with other instruments and the spacecraft are not altered. All of the instrument technology must be at Technology Readiness Level 6 (TRL 6) by the time of the Preliminary Design Review in January 2006.

Mr. Figueroa posed several questions concerning PP and the MSL for the PPAC's consideration. First, what are the PP constraints on landing sites, if there is snowpack or near-surface moisture within  $\pm 30^{\circ}$ latitude? Second what would be the recommendation for sterilization (decontamination) of the MSL and components for the desired latitude band of  $\pm 60^{\circ}$ ? The Mars Exploration Program is currently assuming it will be COSPAR Category IVA. His reply to a question from Dr. Klein on deciding when an improved capability is cost-effectiveness was that the program typically looks for at least an order of magnitude improvement (over the previous mission with similar science capability). For the MSL analytical instruments, the basis is different because they were not at the required TRL for MER. The Program subsequently put more effort into maturing the analytical instrumentation, and he expects this capability to be ready for MSL. Dr. Atlas noted that, because different instruments raise different contamination concerns for a mission with the objective of searching for the building blocks of life, the contamination requirements must evolve as part of instrument selection. Dr. Pieters asked if there was time to use information from MER and MRO in designing MSL. Mr. Figueroa answered that the data from MER will provide "ground truth" to assess the interpretation of data from MGS. Odyssey, and other missions: the MSL instruments will be selected after the MER data have been analyzed. Dr. Noonan agreed but said that the schedule is tight for the data to have optimal impact. Many steps have to fall into place, and spring 2004 will be a busy time for instrument proposers.

The Mars Telecom Orbiter will provide communications relay for MSL, Scout missions to Mars, and the Next Decade of Mars orbital and surface missions. Its optical communications payload will greatly increase the data return capacity for these activities. In introducing the current thinking for the Next Decade of Mars exploration, Mr. Figueroa said the administration has requested that the Next Decade effort be more discovery-driven—using results from missions in place to plan the next steps. A transition will be made from "follow the water" as an exploration approach to "searching for the building blocks of life" (or "follow the carbon"). The four Mars Exploration Pathways for FY 2009–2020 are Search for Evidence of Past Life, Explore Hydrothermal Habitats, Search for Present Life, and Explore Evolution of Mars. The mission sequence reflects a progression of steps along these four pathways, with one mission often providing steps in two or more pathways. Within the current framework, the first sample return mission, the Ground Breaking Mars Sample Return (MSR), would launch in 2013 at the earliest, with samples being returned in 2016. The program will, in the coming year, develop study requirements for a sample receiving facility. Mr. Figueroa and the PPAC discussed sample return capabilities in relation to both sample protection (noncontamination and physical security) and Earth protection, as well as the Program's interest in controlling facility cost. The Astrobiology Field Laboratory (launch circa 2013), as well as the Ground Breaking MSR, would help to prepare for further sample return missions. Dr. Atlas noted the value of

dovetailing the current growth in construction of Biohazard Safety Level 4 (BSL-4) facilities with NASA's need for a planetary sample receiving facility. Mr. Figueroa said that the program hopes to learn from the BSL-4 facilities being built now, to help in designing the sample receiving facility.

Mr. James Campbell of JPL described the status of preparations for engineering studies of a sample receiving facility. They are beginning the process of procuring teams of companies to perform facility systems engineering and cost estimation studies. These teams will use the draft Mars Sample Handling Protocol as the basis for the engineering studies. Responses to a Request for Letter Of Interest, sent to selected companies on May 19, are due in a few weeks. NASA and JPL will take several months to assess the responses, and NASA will also have them assessed by independent outsiders. Dr. Noonan said that these steps appeared to be in line with the PPAC's request that NASA seek preliminary thinking from those with expertise in facilities for biohazard containment. The fall 2003 PPAC meeting would be a good time for a followup briefing on this topic. The PPAC discussed with Mr. Campbell how the companies were selected to receive the Request for Letter of Interest. A working group on facility design, consisting of scientists at the NASA centers, the Centers for Disease Control and Prevention, and the National Institute of Allergy and Infectious Diseases, was polled for names of companies that had performed well in the past. This approach is commonly used for facilities procurements. PPAC members suggested that universities and other institutions that are dealing with the philosophy and basic principles of biohazard safety and containment should be consulted as well. The consensus among PPAC members was that facilities architectural-engineering companies have the expertise to design a facility to meet requirements, but they look to the acquirer of the facility to establish the functional requirements. The PPAC also discussed the role of the draft sample handling protocol. Dr. Noonan summarized by saying that NASA should take the discussion as advice from the PPAC to seek advice more broadly than just from facilities engineering firms. A mechanism for continuing interaction between the PPAC and the Mars Exploration Program on this issue was also discussed.

#### Mars Simulation Studies of Microbial Survival

Dr. Andrew Schuerger described his continuing studies and interim results on microbial survival in simulated martian atmospheric and surface conditions. The broad purpose is to look at scenarios for survival of bacteria on landed spacecraft or vehicles in martian environments. The survival rate for *Bacillus subtilis* endospores decreases substantially after just a few hours of exposure to the vacuum of space and decays along a hyperbolic curve. For a 200-day transit to Mars, the survival rate for an endospore population could be as low as 25 percent. The damage to non–spore-forming bacteria in space vacuum is significantly greater, typically showing 2 to 3 orders of magnitude reduction in just a few hours. In response to a question from Dr. Cavanaugh, Dr. Schuerger said that only 2–3 percent, to at most 5 percent, of the microorganisms present are picked up by culturability tests. Dr. Cavanaugh noted that extremophiles in particular cannot be cultured by the standard methods. Therefore, a list of culturable bacteria found on spacecraft does not address questions of whether extremophiles may be present or what microorganisms might survive martian temperatures and atmospheric conditions (lack of oxygen).

Dr. Schuerger's simulation studies to date have focused on Bacillus subtilis endospore survival in the ultraviolet (UV) environment of sun-exposed surfaces on Mars. The Mars Electrostatic Chamber in the Materials Sciences Laboratory at KSC was used to simulate the UV exposure, as well as the temperature, pressure, and gas composition of a martian environment. One difference is that the light used does not include the infrared component of martian sunlight, which would have damaged the UV filters. The unfiltered source delivers a Mars solar constant light equivalent, and filters were used to represent different atmospheric attenuation conditions corresponding to specific optical depths (tau values). The bacterial spore cultures were exposed on aluminum coupons, and a standard Most Probable Number assay technique was used to determine population survival rates. In the tests, one environmental parameter at a time was varied, with the others held at the Mars-normal conditions. The tau values ranged from 0.1 (top of the atmosphere) to 3.5 (equivalent to a global dust storm on Mars). Even at tau = 3.5 attenuation, biocidal conditions (survival rate = 1 percent) occur with 60 minutes of exposure. Neither gas composition nor temperature affected survival rate relative to UV exposure at different tau values. In tests comparing spores not exposed to UV (an "inside surface" population) and UV-exposed populations ("outside surface" population), about 20 percent of the nonexposed endospores appear to die off under any of the conditions tested. Settling of dust particles onto bacterial monolayers did not protect the endospores. Although a 0.5-

mm contiguous layer preserves the spore population, dust treatments representing actual atmospheric settling of dust particles did not. Dr. Schuerger noted that the older studies on spore survival in martian conditions did not use UV sources that represent the full UV spectrum at the martian surface. He showed LD<sub>90</sub> (lethal dose for 90 percent of a population) values for spore-forming and non-spore-forming bacteria. His conclusion is that a lethal UV dosage accumulates quickly for exposed spores, even for spores of the most UV-resistant bacteria. However, survival rates vary for different materials (astro quartz and graphite retain detectable populations after 1 hour of UV irradiation while metals do not). Dr. Schuerger thinks that the spore inoculations on quartz and graphite are not drying down as monolayers. Microdrops form and contract, concentrating the spores in thick aggregates (multilayers rather than a monolayer). Debris from indeterminate sources may cover spores on porous materials. Spacecraft surface characteristics can affect the attachment and survival of microorganisms under simulated martian UV irradiation. In addition, the UV irradiance on Mars varies considerably with the solar zenith angle. Studies of direct UV and diffuse UV indicate that, even under global dust storm conditions, the UV dosage is close to 25 percent of Mars-normal conditions. Dr. Schuerger answered questions from the PPAC members on the relation of his results to those in the older literature and in newer studies done using methods similar to his. While his studies to date have been on fully exposed surfaces, he will next investigate survival under buried and other protected conditions. The PPAC asked additional questions related to density of populations, presence of multilayers or biofilms versus monolayers of spores, effects of dust coatings, and survival rates of life forms other than bacteria. Dr. Schuerger iterated the point that much of the older literature does not simulate Mars-normal pressure, temperature, gas composition, and UV.

#### Friday, May 30, 2003

#### Viking Planetary Protection and Current Standards

Dr. Pericles Stabekis described the derivation of PP standards for the Viking missions, how the Viking mission team achieved them, and how those standards became the current standard. The governing policy circa 1969 was a COSPAR/NASA quantitative policy. The basic objective for planetary quarantine of Mars and other planets set a probability of not more than 1 x  $10^3$  that a planet would be contaminated during the period of biological exploration, which was assumed to extend for 20 years, ending in 1988. COSPAR also recommended that members provide a report on sterilization procedures and residual bioburden computations within 6 months of each launch. The total probability of contamination was then allocated among launching nations. Each launching nation gave an allocation to its planetary missions. NASA allocated 2 x  $10^{-4}$  to the two Viking missions. The Viking 1 allocation for the lander was  $3.25 \times 10^{-5}$ . A probability of spore survival was derived through a logarithmic death rate model,  $P_s = 10^{-t/D}$ , in which  $D_T$ , the D-value for a fixed temperature T, was determined using *Bacillus subtilis var. Niger* as the standard organism.

Even before the Viking 1 launch, a microbial subpopulation was known to exist that had resistance to heat far greater than the standard organism did. The Viking sterilization cycle reduced populations of these microorganisms by only a decade (to 10 percent of the starting population). The Viking bioburden report to COSPAR was recast to include an evaluation based on the presence of hardier organisms. The final probability of contamination for Viking 1, using an amended  $P_s$  value to reflect the hardy spore population, was calculated as  $3.08 \times 10^{-5}$ .

The Viking computations assumed a probability of growth of surviving spores of 1 x 10<sup>-6</sup>, an estimate proposed by the SSB. After Viking, the SSB became increasingly reluctant to propose such estimates because of the uncertainties involved. Instead, the SSB recommended a policy of categories for procedures and cleanliness standards to be achieved. NASA accepted this policy and proposed it to COSPAR in 1984. In effect, the policy provides guidelines for implementation of cleanliness procedures, while the top level of COSPAR policy remains a quantitative standard. In its 1992 report on protecting Mars, the SSB recommended revising Category III and dividing the original Category IV into IVa and IVb. COSPAR adopted the SSB recommendations in 1994. The NASA PP program then issued specifications on spore bioburden for landers using these refined categories. The specifications include ceilings on both surface concentration bioburden and total bioburden. In 2002, COSPAR reaffirmed the 1994 policy in a document that integrated all the requirements. A new Category IVc was introduced for landing in martian "special regions" if the lander does not have life detection experiments. Special regions are those within which

terrestrial organisms are likely to propagate or which are interpreted to have a high potential for existence of martian life forms. In practice, this means regions where liquid water is present or may occur. Dr. Stabekis concluded with six points for the PPAC to consider. (1) The current cleanliness levels for Mars landers derive from the Viking levels, which depended primarily on estimates of two parameters, the probability of contamination and the probability of growth. (2) The post-Viking changes to policy eliminated the top-level probability of contamination allocations and the need for a probability of growth estimate. (3) The current PP specifications for probability of impact on Mars are consistent with a probability of contamination of  $10^{-2}$ . (4) The definition of martian special regions implies a probability of growth far higher than the Viking estimate of  $10^{-6}$ . (5) The assay methods for cleanliness, based on culturability, may be outdated. (6) The Mars lander requirements, particularly those for Category IVc, need to be revisited to reflect the new data on Mars and on microorganism survivability.

In response to a question from Dr. Noonan, Dr. Stabekis said that mission cost is the factor always weighed against just going to a Viking post-sterilization standard of cleanliness for any Mars lander. At present, dry heat is the only method accepted for "sterilization" (highest level of decontamination), although hydrogen peroxide treatment is being considered as an alternative. The key problem in examining alternative sterilization methods is compatibility of spacecraft/lander components with the new method. The PPAC discussed the factual basis for the cost of a higher standard of decontamination and whether modern electronic components could withstand the temperatures attained in the Viking heat sterilization procedure. Dr. Noonan concluded that there appear to be two directions to take: either assume that heat decontamination is sufficient and appropriate or that another method or technology, such as hydrogen peroxide or irradiation, is needed as an alternative or complementary process to decontaminate the spacecraft/lander. In response to Dr. Griner's question about the COSPAR category for potentially life-supporting regions on bodies other than Mars, Dr. Rummel said that all of Europa, for example, is considered a "special region" for purposes of the COSPAR classification.

#### Solar System Exploration Program Status

Dr. Colleen Hartman, Director of the Solar System Exploration Division (SSE), updated the PPAC on PP issues related to missions and plans in SSE. The Galileo spacecraft was in orbit around Jupiter since December 1995. In January 2001, it was placed in a trajectory for impact on Jupiter in September 2003. This disposition is in the spirit of the 2000 SSB report on preventing contamination of Europa and complies with the recommendation of the SSB's Committee on Planetary and Lunar Exploration. Stardust, the fourth mission in the Discovery series, has a sample return of comet and interstellar dust particles from the vicinity of comet Wild 2. The sample return capsule will deploy parachutes for a soft landing in the Utah desert. The sample will be curated at the Johnson Space Center (JSC) and SSE has allocated \$400,000 for an upgraded cleanroom to house the sample. Dr. Hartman's briefing package contained additional data on the entry, landing, and post-landing sample handling procedures. The FY 04 budget request for SSE includes an Astrobiology line item, which includes support for research in the PP-relevant areas of life in extreme environments and instrumentation for detecting signs of life. This budget item provides \$60 million more for astrobiology than was in the FY 03 budget. The Astrobiology program provides a mechanism for funding PP research through open competition and joint review panels. The first competition is likely to be announced in January 2004. New techniques for monitoring and reducing spacecraft contamination will be included in the program. Dr. Hartman said that, if FY 03 funds are available, she would like to issue a NASA Research Announcement (NRA) for research related to outer planetary exploration. The NRA would include materials research related to techniques to reduce forward contamination.

The Genesis mission involves return in September 2004 of a sample of solar wind particles. A Memorandum for the Record stating that Genesis is classified as an unrestricted Earth return was signed in July 2001, and Genesis compliance with the National Environmental Protection Act (NEPA) was completed in August 2001. A sealed canister will be delivered to the sample curation facility, which has two Class 10 cleanrooms (i.e., a maximum of 10 particles per cubic foot), which were funded by the project. Dr. Hartman's briefing contains backup slides with details of the potential environmental impacts of sample return and recovery operations. Dr. Hartman reviewed the battery temperature problem with Genesis and commented on potential consequences for the mission.

The New Horizons/Pluto-Kuiper Belt mission will be the first in the New Frontiers program. Missions in this program are capped at \$650 million. The President's FY 04 budget request includes a \$210 million increase for the program in FY 05. A major issue is that the budget of \$110 million for New Horizons is less than requested, and the launch date is in FY 06. Dr. Hartman discussed how this relates to other issues in ensuring that the mission progresses successfully. New Frontiers also has funding for a new mission start in FY 03, and each of the four highly ranked mission types in the Announcement of Opportunity (AO) has PP implications. These missions are (1) South Pole (Mars) Aitken Base Sample Return, (2) Jupiter Polar Orbiter with Probes, (3) Venus In-Situ Explorer, and (4) Comet Surface Sample Return. Dr. Hartman reviewed the constraints on New Frontiers missions written into the authorizing legislation.

What had been the Nuclear Systems Initiative (NSI) in the FY 03 budget has been brought into the new Project Prometheus. The science for Project Prometheus missions is in SSE, while the technology is under the Project Prometheus technology program. The program for radioisotope power systems (RPSs), which are essential for the Discovery missions, is now part of Project Prometheus. For the other two parts of Project Prometheus, nuclear electric power and nuclear electric propulsion (of spacecraft), a Nuclear Concept Definition Team, now headed by Dr. Eugene Levy, will advise Dr. Hartman. The central question it will address is, "What kinds of missions and scientific research in deep space would be enabled by 100–250 kWe of available power?" Dr. Hartman provided some comparisons of the power and energy capabilities of missions with reactor power versus solar and/or chemical (battery) sources.

The JIMO mission is currently an orbiter mission, although some in the science community are thinking about the possibility of including landers. Dr. Hartman thinks that lander concepts are more likely to be relevant to subsequent missions. There would be PP issues associated with landers, as well as PP issues if nuclear electric propulsion is the only power source on board and a reactor fails during a sensitive part of orbiter maneuvers, such as during a spiral toward or away from a body. Dr. Hartman summarized the science-related activities scheduled for JIMO. She asked the PPAC for input on PP issues in future SSE missions and for PP research topics that could be included in the Astrobiology research program. Dr. Noonan added that the PPAC will need to consider the PP implications of nuclear electric power and propulsion, including determining who in NASA is addressing the implications of reactors landing on planetary bodies.

#### Jupiter Icy Moons Orbiter Mission Status/Plans

Mr. Alan Newhouse, Director of Project Prometheus, briefed the PPAC on the plans for developing nuclear electric power and propulsion technologies under Project Prometheus and the implications for the JIMO mission. One part of Project Prometheus is to revitalize the radioisotope thermoelectric generator (RTG) program and establish plutonium sources for RTGs and other RPSs. Within the Project Prometheus nuclear power/propulsion program, the three aspects being pursued are research (through the Department of Energy) on appropriate nuclear fission reactors, power conversion research and electric propulsion research. Mr. Newhouse discussed the advantages of the much greater energy and power levels for science missions. He described the value of nuclear electric power for science instruments, science data return, and the time available for study after a mission is on station, as well as differences in maneuver capabilities between current propulsion regimes and nuclear electric propulsion. Indirect benefits of the program include education and spin-off technology. In his discussion of matching the type of power system to the planetary destination, he included RPS capability for surface landers on all planetary missions.

Mr. Newhouse described the JIMO mission as providing a clear science rationale for proceeding with nuclear electric power/propulsion technology. He reviewed the past and current systems that incorporate RTGs and the PP considerations relevant to them. For example, RHUs (radioisotope heating unit) are used to keep components from getting too cold to operate. He also discussed the PP considerations for JIMO. The current thinking about a JIMO launch date places it in 2011. His program is considering ways to decrease the flight time to Jupiter to 5–6 years, rather than arriving in 2020, as in the current scenario. In the following discussion of when input from PPAC would be useful to his program, Mr. Newhouse said that it was important to know if anything special will be needed to address PP issues. He also discussed with the PPAC the risk communication effort to determine how the public views the prospects of nuclear-powered missions. The PPAC discussed which aspects of nuclear-power issues are within the committee's purview.

#### NRC Mars Forward Contamination Study

Ms. Pamela Whitney, a Senior Program Officer on the staff of the SSB, described the new NRC study on preventing forward contamination of Mars. This study will update the 1992 study, *Biological Contamination of Mars: Issues and Recommendations*. The tasks include: (1) assessing and recommending levels of cleanliness and sterilization required to prevent forward contamination of Mars, (2) reviewing methods to achieve and measure the appropriate levels of cleanliness and recommending alternatives in light of recent science and technology, and (3) identifying scientific investigations that should be accomplished to reduce the uncertainty in the above assessments. The NRC has just received funding for the study from NASA. The staff is currently selecting a nominee for committee chair, who will then aid the staff in preparing a nomination slate for the rest of the committee. The committee will require expertise in Mars science, extremophiles, biochemistry, microbial ecology, and spacecraft systems. Ms. Whitney agreed with Dr. Noonan's suggestion that individual PPAC members consider potential nominees in the disciplines relevant to the Statement of Task and send their suggestions to Ms. Whitney at the NRC. The committee is expected to begin meeting early in the fall of 2003. There will be three meetings, one of which will be a workshop, during the 15-month study schedule.

Dr. Cavanaugh asked about the extent of overlap in scope between the new NRC committee and the PPAC. Dr. Rummel replied that the two bodies overlap in scope, with respect to Mars forward contamination issues, but differ in the mechanism, timing, and forms of advice to NASA. The NRC study committees provide periodic inputs (several years apart), which affect the NASA policy framework. In addition the National Academy of Sciences [of which the NRC is the operating arm] is the U.S. member of COSPAR. The PPAC provides more routine, quick-response advice to PP activities in NASA programs, within the overall NASA policy framework. Dr. Noonan added that the 1992 SSB report does not reflect the understanding of Mars environments that has emerged since then, so this update will be useful for NASA and the PPAC. She added that, after the chair of the new committee is appointed, Dr. Rummel and she will plan to meet with that chair to agree on ways to coordinate and communicate between the PPAC and the NRC committee.

#### Upcoming Mars Planetary Protection Issues (Closed)

The PPAC entered a previously announced closed session to discuss PP issues that could arise with Mars missions proposed as part of the Scout mission competition. Dr. Rummel began by stating the conflict-of-interest requirements for attending the closed session. All attendees had submitted a nondisclosure and conflict of interest agreement. Dr. Rummel described the four Scout missions being considered for selection. The missions have just completed the concept study phase and have submitted their studies. NASA anticipates selecting only one mission for Phase B. Dr. Rummel reviewed the COSPAR PP mission categories. He then discussed the COSPAR categorization for the four candidate missions, the rationale for how each mission is categorized, and the PP elements each mission team had proposed to meet the assigned category requirements. The PPAC then discussed PP issues of each candidate mission. Dr. Noonan asked whether PP considerations would be explicitly brought up during the peer review. Dr. Rummel explained that he performs a PP review of the proposals, then the technical management cost group will assess whether each project team can accomplish what it claims within its proposed budget. The science review panel will consider how the PP requirements might affect the science objectives of the mission.

#### Tour of Kennedy Space Center and Cape Canaveral Air Force Station

After the closed session, Dr. Rummel reviewed the agenda for the PPAC's afternoon tour of KSC and CCAFS. After entry and security screening at the KSC main entrance, the group proceeded by bus to the processing facility where the MER-B spacecraft is being prepared. The facility was closed to visitors for a spin test of the spacecraft, so the PPAC observed the spacecraft via remote camera from the NASA mission monitoring facility (Hangar AE). At Hangar AE, they also observed the Space Infra-Red Telescope Facility, which is being held in a cleanroom awaiting launch. The PPAC visited the launch area for MER-A, which is located on the CCAFS. The tour also stopped briefly at the Shuttle launch site before ending at the Apollo/Saturn V Center.

#### Saturday, May 31, 2003

#### **Committee Discussion**

After a review of the day's agenda, Dr. Noonan asked the members to email her any comments on the structure of the meeting, particularly the amount of time available for discussion. The PPAC then discussed results from a Mars sample return special study group on evaluation of post-collection temperature effects on sample properties. A basic reference on this topic is a NASA technical memorandum prepared by. J. Gooding in 1990: *Scientific Guidelines for Preservation of Samples Collected from Mars.* NASA Technical Memorandum 4184. This document lists mineralogical properties that would and would not be changed by temperatures of 200 °C. Dr. Atlas pointed out that sterilization depends on the combination of time and temperature. Decimal reduction times (death rates) for a known time and temperature can therefore be used to estimate temperature-time combinations that will give a specified reduction from initial populations. The PPAC discussed how to apply what is known about death rates of known organisms to unknown conditions (including unknown organisms and/or unknown concentrations and environmental conditions).

Dr. Noonan asked for the sense of the committee on the issue of public announcement of the availability of the draft Mars Sample Handling Protocol. The members agreed that the next letter from PPAC to the NASA Administrator should state the PPAC's strong desire to have a Federal Register notice of the draft Protocol, if it has not already been published there. The letter should also express the PPAC's willingness to work with the NASA Office of General Counsel if that office has issues or concerns regarding a notice. The PPAC next discussed current and forthcoming NRAs with PP-related research topics. The members agreed that the letter from PPAC should express gratitude to NASA for the Mars Exploration Program's recent announcement on PP-related research and for the future possibility of one or more Astrobiology NRAs with PP topics. The letter should also note the value for NASA in supporting these research topics. Dr. Atlas added that NASA should support research to reduce the burden of false positives in molecular methods for monitoring microbial populations, not just research on methods to kill organisms. Dr. Cavanaugh raised the issue that molecular methods are needed that distinguish between the molecular signatures of still viable organisms from the signatures of biomolecules that remain even after the organisms that produce or contain them are dead. Another issue is that NASA's standard testing for contamination still depends on culturation methods, but less than 1 percent of viable organisms in many environments are culturable. Dr. Noonan asked the members to think of other issues for the letter to NASA, with the discussion to continue later in the day.

#### MER Planetary Protection Compliance Status

Ms. Laura Newlin, PP lead from the JPL team for the MER project described, the compliance procedures and status for the two MER rovers, which must meet COSPAR Category IVa requirements. Because the backshell will not heat to 500 °C for the 0.5 s during entry required for exemption from launch bioburden assessment, the backshell, cruise stage, fairing, and other parts required cleaning and assaying. One difference from some earlier Mars missions has been that PP planning and implementation has been fully integrated from the beginning of the project, rather than having to play catch-up late in the implementation phase. Design requirements included probabilities of events, bioburden limits, and a listing of all organic materials and their masses. From these base design requirements, the team derived additional design requirements for the hardware, such as compatibility with damp swab assay sampling, compatibility with either alcohol wiping or dry heating for decontamination, and sealing of enclosed electronic modules with high efficiency particulate air (HEPA) filters. Implementation approaches were a mixture of dry heat microbial reduction, isopropyl alcohol wiping, isolation with HEPA filters, and sterilization by contact with hydrazine propellant. Ms. Newlin showed graphics of the lander and components to illustrate which decontamination methods were used for various parts of the lander. In reply to a PPAC question about the bioburden in components sealed by HEPA filters, Dr. Rummel explained that there are requirements during assembly of the components to reduce contamination, but they are sealed off because they cannot be assayed after assembly. The cruise stage does not have to meet a bioburden limit because it is assumed to reach 500 °C before it crashes on the martian surface.

## MRO Planetary Protection Status

Jack Barengoltz of JPL discussed the status of PP work for the MRO. The project team initially tried to meet the PP requirements using a probability of impact computation, as NASA has done in the past for

Mars orbiters. But the very low science orbit for MRO meant there was a significant probability of impact (greater than 1 percent) early in the orbiter's lifetime. The project therefore moved to a total bioburden approach to meet the PP requirements, but the computations of bioburden, assuming standard assembly and cleaning approaches, do not meet the 5 x 10<sup>5</sup> ceiling on spores. The project has negotiated with NASA for an approach based on the bioburden in just those parts of the orbiter that would reach the surface of Mars, given a failure in orbit, without reaching 500 °C for at least 0.5 s. Thus, the bioburden estimation now depends on the results of the Breakup and Burnup analysis. Lockheed-Martin-Albuquerque was selected to do the analysis, which assumed an aerodynamically stable configuration during a breakup trajectory. When the results of the analysis were sent to the NASA PP Officer for review, questions were raised about the consequences of a tumbling entry and about possible cross-contamination. The tumbling analysis has now been completed, and subsequent work supports the view that tumbling entry is not possible. The crosscontamination analysis estimated fewer than 8.4 x 10<sup>4</sup> spores deposited on critical surfaces during launch, but additional Breakup and Burnup analysis shows that all these surfaces would be ablated or sterilized during entry prior to impact. Mr. Barengoltz presented the PPO reviewer comments in detail and the project responses to address them. The launch is scheduled for August 2005. The expanded Breakup and Burnup analysis will be completed this summer. The PPAC discussed the problems that have arisen with spore contamination on the MER fairings and what could be done in the future to ensure that bioburden is controlled during manufacture of fairing materials.

#### Communication Issues in Planetary Protection

Margaret Race, from the SETI Institute, began the presentation on risk communication issues in PP, followed by Linda Billings, also with the SETI Institute. In her past work on PP and risk communication, Ms. Race distinguished three phases. From 1992 to 1994, she focused on research to understand risk perceptions and risk communication needs related to planetary protection, in the context of public decision making. Potential public reactions to returning Mars samples to Earth were compared with similar cases that generated public concern, including the Apollo returns of samples from the Moon, genetic engineering (Ice Minus), the RTG-equipped Cassini mission, and the Galileo mission. These comparisons suggested that sample return would be viewed as high risk, like genetic engineering. Success in gaining public acceptance would depend on (1) having solid, current science and technology to support the mission; (2) integrating PP with mission planning and implementation from the start (not an add-on); (3) staying aware of legal ambiguities and issues; (4) ensuring interagency coordination; and (5) taking a direct approach in addressing societal concerns. The Internet-based protests against the Cassini mission showed the Internet to be an emerging source of information (and misinformation), opinion, and public reaction to science activities perceived as carrying high societal risks.

The second phase of research, from 1995 to 1999, focused on communicating with expert audiences about substantive issues in PP and comparing expert perceptions with those of lay audiences, to understand differences in perceptions of risk in space exploration. Survey-based research, focus groups, and Internet website analyses were used. The outreach to expert audiences was intended to deliver two messages: PP is an essential part of space exploration, not a public relations sidelight, and experts need to understand the societal context and concerns with which lay audiences view space exploration.

From 2000 to the present, Ms. Race's work has begun to include proactive outreach. The technical context for this phase includes the drafting of a Mars Sample Handling Protocol, refining PP control methods, planning for sample return environmental impact statements under NEPA, and the relation between PP and human missions. The risk communication effort during this period has included continued publication of articles about PP plans for missions in diverse scientific and technical journals. A consistent message about PP emerged with the "All the Planets, All the Time" motif. Numerous activities have aimed at translating PP issues for general audiences, using a variety of forums (magazines like *Discover, Astronomy*, and *The Planetary Report*; shows on The Discovery Channel, BBC, and NPR; etc.). Another part of the effort has been to monitor pubic concerns and responses to large-scale societal issues with a science context, including bioterrorism, weapons of mass destruction, emerging diseases, nuclear issues, and space exploration.

Linda Billings discussed the next phase of PP risk communications, which will focus on increasing the tempo of communicating about PP. NASA needs to be prepared for public response, and it is best to be

proactive. The context is one of growing public involvement in scientific research decisions (e.g., constraints on stem cell research, genetic engineering). The sorts of communication that are needed include general information, communications with target groups (government overseers, skeptics, opponents, mass media, etc.), and disaster/emergency communications. Ms. Billings concluded that what NASA needs in its PP risk communications are a participatory/interactive model, a story that is clear and concise, trust (in institutions and in science; also in individuals) as the top priority, and research to characterize and cultivate the multiple audiences. For this phase, Ms. Billings is currently doing qualitative research on science communication in journalism, public affairs, risk communication, and government and nonprofit agencies. Next steps include drafting a communication strategy for PP policy, activities, and events

Dr. Kerridge asked whether the interface on PP with the public should be a separate entity, independent of NASA, rather than an entity perceived as part of NASA or under its control. This led to a general discussion of what kind of entity would have the greatest public trust on PP issues and how NASA perspectives can best be advocated before the public. Dr. Atlas suggested that members of the public should be incorporated in the entity, such as has been done on NIH's Recombinant Advisory Committee.

#### Planetary Protection Training Course

Dr. Gary Coulter described the training course he is preparing, titled "Planetary Protection: A Course for Practitioners." As the target audience, practitioners include those responsible for implementing PP procedures anytime from design through launch, operations, and sample recovery. Those who implement or are charged with ensuring the implementation of PP policies and procedures are also practitioners. The desired outcome of the course is to give all personnel a more detailed and comprehensive understanding of PP policies and procedures, including the ways in which individual activities must be coordinated to implement a successful PP program. The minimum class size is 8, the maximum is 20. There will be a fee for taking the course, which will be revenue-neutral with respect to the Planetary Protection Program, and attendees will be responsible for their own transportation, lodging, and meals. The three initial venues for the course are the Center for Marine Biotechnology, Baltimore; the Wrigley Marine Science Center, Catalina Island, California; and a European venue at ESTEC in the Netherlands. The first session will be in the fall of 2003; the second in the fall or winter of 2003-2004. The European session may be in the spring of 2004. The course will be taught by PP practitioners from NASA, JPL, the SETI Institute, and other programs in NASA and other institutions. Dr. Coulter presented the draft outline for the course and asked PPAC members to send him comments on it, if they wished. He reported substantial enthusiasm from those who have been contacted about having members of their group attend the course. Next steps in implementation are to develop a trifold brochure and electronic information materials, obtain commitments to attend, and finalize the course logistics.

Dr. Cavanaugh asked that the issue of nonculturable microbes be covered in the course. Dr. Noonan suggested that it should communicate why new monitoring and decontamination methods are needed and what the alternatives may be.

#### Mars Program Education and Public Outreach (E/PO)

Michelle Viotti, Mars Public Engagement Manager, described her public engagement work and its relation to PP and risk communication. She defined public engagement as covering all products and activities that are intended to reach the general public. In addition, public engagement is two-directional, with active collection and reception of public feedback. The central messages are that NASA is listening and responding, not just reaching out with information. A good example of this engagement approach is the Mars Student Imaging Project, which involves K-12 students in archival image investigations, distance learning, and on-site investigations. Another example is the Mars Robotics Education Program for students. The Florida: Gateway to Mars program fosters risk communication with the KSC local community about nuclear materials risks. Ms. Viotti noted that the latter program has not yet addressed either Mars forward contamination or sample return issues. The three broad categories for Public Engagement are formal education (classroom learning tied to education standards), informal education (such as facilitated learning in museums and other non-academic settings), and public outreach in general "pass through" events such as lectures to a broad range of audiences.

Ms. Viotti defined "risk communications" as a coordinated approach for communicating with the public media, educators, students, legislators, and governmental bodies—about NASA missions that: (1) use radioisotope power systems, radioisotope heating, or other nuclear elements; (2) return samples from bodies in space to Earth; (3) conduct experiments in pristine Earth or space environments; and/or (4) have environmental safety issues that may be of greater than ordinary concern to the public. Within NASA, Public Engagement and Risk Communications activities are led by different offices. A subset of Public Engagement products and activities are specifically designed in coordination with the JPL Risk Communications Coordinator and NASA HQ to inform and educate the public about topics of potential interest and concern. Ms. Viotti stressed that responding to public interest will not mean being reactive. Rather, it means a proactive strategy for understanding and addressing public issues and concerns; a commitment to two-directional, open, and honest communications at all times; and a commitment to providing an easily accessed, respectful environment for dialogue. She summarized the results of the Mars Exploration Survey, which was a self-administered Internet-based survey designed to gather respondents' views using broad, non-leading questions. Among the notable trends from the 537 respondents were frequent responses citing "fears for Earth" and the possibility of Mars as a new or second "home," in answer to the questions. "Why do you think we should explore Mars?" and "How do you think Mars exploration benefits the world?" Also, the public tends to see the nuclear issue as part of 'planetary protection."

As areas where improvement is needed, Ms. Viotti had three suggestions. More attention should be paid to internal communications. More coordination across programs (beyond just the Mars Program) is needed to share risk communications issues. Those speaking for or about a particular program or project need to think in terms of, and be familiar with, the overall NASA effort, not just one area. As areas where NASA has great strength, she said that a proactive strategy on risk communications is being implemented now, many years in advance of missions that could generate public reaction, such as MSL or Mars sample return missions. Another strength she sees is that the program is genuinely committed to being open and receptive.

#### Committee Discussion

Dr. Atlas drafted the following questions and comments for the PPAC's consideration. (1) Are electronics more resistant to radiation than to dry heat? Perhaps UV radiation can be used for sterilization, instead of heat? Radiation could also destroy materials used for molecular signatures. (2) What about higher temperatures and shorter times for decontamination (analogy: food sterilization)? What about alternative methods of sterilization (analogy: mail sterilization)? Also consider targeted approaches, e.g., using intrinsic fluorescence. (3) Conflicting considerations regarding where bioburden may occur: Surfaces may be more highly contaminated, but the UV killing rads are greatest for exposed surfaces. Internal contamination may be of greater concern; the likelihood of contamination may be lower, but microorganisms are less likely to be killed by radiation. (4) Are there methods that destroy DNA/RNA and not just "sterilize" [kill microorganisms]? Dr. Rummel suggested that further consideration of these issues could be a future PPAC activity. The members discussed UV irradiation as a way to support cleaning and destruction of bioburden throughout fabrication and assembly of components. They also raised the issue of the stage in production and assembly at which bioburden reduction should become an objective, or even a requirement, for materials qualification.

During lunch, dates for the next meeting were discussed. The window of December 15–17 was agreed upon as the best time for a two-day meeting.

#### Applied Molecular Methods for Planetary Protection

Dr. Norman R. Wainwright, Marine Biological Laboratory, presented his work to date on a "Limulus Amebocyte Lysate (LAL) Test for Planetary Protection." The presentation covered progress and problems in seeking a near–real-time test for detection of microbes, specifically the use of amplified enzyme cascades for microbial detection. The project team coupled the last step in a Limulus coagulation cascade (Limulus is the horseshoe crab) with a chromogenic substrate, *p*-nitroaniline, which turns yellow in response to the cascade. The basic test has been used in the pharmaceutical industry for a number of years to qualify drugs for injection. The endotoxin units (EU) used in Dr. Wainwright's charts are roughly equivalent to 100 picograms of lipopolysaccharide (LPS), and the sensitivity of the assay is about  $10^{-12}$  g of

LPS. As a model for using the test on a returned sample, Dr. Wainwright presented data from tests of samples of lunar material. The assay distinguished clearly between dust from astronauts' space suits, which had been contaminated prior to sample collection, and the uncontaminated lunar materials. He also presented data comparing the LAL test with a standard plate culture assay (without heat shock prior to culturing), using samples from spacecraft materials and areas around spacecraft assembly cleanrooms. PPAC members asked about the response of the assay to different types of microbes (e.g., gram negative and gram positive bacteria, yeasts and molds). Dr. Wainwright noted that he is working on another complement cascade test, from a different crab species, which will be sensitive specifically to gram positive cell wall materials.

As future work, Dr. Wainwright described the Quality Assurance Test Plan for both the LAL and ATP assays. The objective of the plan is to provide the data necessary to support statistical evaluations equivalent to the Viking standard of decontamination. Both assays will be tested for reliability, limits of detection, and feasibility as a portable assay method. A portable (handheld) system for the LAL assay has been developed in conjunction with the company that makes the LPS reagent. Dr. Wainwright presented data comparing the laboratory-based LAL "standard method" with the portable test system. Work in progress includes sampling of the MER spacecraft by both LAL and standard methods to establish comparisons and relevance of the EU scale, developing the test for gram positive sensitivity, continuing the work on miniaturization for portability and field analysis, and adapting the LAL assay for use in life detection technology for astrobiology experiments.

Dr. Kasthuri Venkateswaran of JPL reported on his group's study of ATP as a biomarker in enumerating total and viable microbial burdens of spacecraft and associated environments. The universality of this chemical assay stems from ATP being an energy source used by all known life forms. It is also a fairly stable molecule linked to evidence of life. The basic ATP-bioluminescence assay he used is a commercially available test. The test kit available from Kikkoman Corp. differentiates free extra-cellular ATP (ATP from dead cells, etc.) from intracellular ATP (ATP in viable microbes). Calibration curves for E. coli and Bacillus subtilis show that the ATP bioluminescence assay correlates with cell number (CFU/ml) over the range from 10 to  $10^7$  CFU/ml for both gram negative and gram positive bacteria. Because endospores contain AMP rather than ATP, the assay is not sensitive to spore populations. Dr. Venkateswaran answered questions from PPAC members on the swabbing method and the use of water rather than buffered solution for swabbing. He also discussed comparison of colony counts versus intracellular ATP, as the amount of ATP depends on the vegetative state of the bacteria. In general, the ATP bioluminescence assay gave cell numbers 1 to 2 orders of magnitude higher than the culture assay. An analysis of samples categorized according to the relative results of the ATP assay and standard cultures found that samples where the ATP count was much higher than the culture method had more yeast, fungi, and gram positive species. Samples with an ATP count but no cultivable organisms typically contained anaerobes, thermophilic bacteria, or oligotrophs. The ATP assay gave fewer counts than the culture assay when spore-forming species were prevalent in the sample. Dr. Venkateswaran said the increase in microbial contamination found by the ATP assay, compared with the standard culture assay, is most likely due to the presence of viable but noncultivable microorganisms. Although the ATP population estimates correlated with whether a facility was classified (as a cleanroom/area) or not, there was no difference between various classes of "classified" cleanrooms.

For alignment with Mars Program goals in the near term, the ATP assay enables rapid identification of likely contamination "hot spots" on exposed surfaces. Dr. Venkateswaran favors amending the standard procedure for microbiological examination of space hardware (NHB 5340.1C) to allow use of the assay for that purpose. In the longer term, he sees the assay enabling a more complete assessment of bioburden contamination for future missions, as well as providing a useful bioburden assessment tool for Mars sample return and sample handling. He closed with a summary of ATP assay data taken from MER swabs, both before the spacecraft left JP and after it arrived at KSC. His summary points were (1) ATP is a useful biomarker to enumerate viable microbial burden; (2) ATP can be used to estimate viable non-cultivable microbial burden; (3) Although ATP is not a spore biomarker, it can be used to estimate overall microbial burden of the spacecraft and associated environments; and (5) the ATP-based enumerating system is rapid, simple, and easy to perform; it can be miniaturized and is field deployable and commercially available.

Dr. Venkateswaran ended with a discussion of work in characterizing microbes isolated from spacecraft surfaces that are resistant to UV, have unusual spore structures that protect them, or produce bioactive compounds (enzymes or glycoproteins). He is studying methods of spore quantification in real time that use AMP, dipliclonic acid (a spore core component), or spore-specific genetic markers. One application for the study's results on enumerating specific types of microbes is a microarray ("phylogenetic biochip") for quantifying multiple microbial populations at high spatial and temporal resolution.

#### Discussion on Applications of Molecular Methods as Standards

Dr. Rummel summarized the status of molecular methods for detecting microorganisms. Issues of comparability among methods and of comparability with the standard culturation method still need to be resolved. The NASA PP Office has held workshops on translating the current standard to molecular methods.

Dr. Levy asked whether the agar cultivation standard should still be considered relevant as a standard for martian life forms. Dr. Pieters asked whether raising the standard for decontamination (to prevent forward contamination) too high might make planetary exploration missions prohibitively expensive. The members discussed different approaches to expanding testing methods and standards to a multimethod approach, rather than relying on a single limited approach such as cultivable spores. They also discussed variability in microbe populations in different environments. Dr. Noonan closed the discussion by noting the relevance of these issues to PPAC's charge, as well as to the NRC study that is beginning. She said that, at the next meeting, the PPAC should consider how to assess decontamination techniques. The meeting adjourned at 3:00 p.m.

PPAC Meeting May 29-31, 2003
Appendix A

# PLANETARY PROTECTION ADVISORY COMMITTEE (PPAC)

Sea Oats Room, Hilton Cocoa Beach Cocoa Beach, Florida May 29–31, 2003

### **AGENDA**

Day 1—Thursday, 29 May 2003

6:30pm	Welcome and Meeting Overview	Norine Noonan/John Rummel		
6:45pm	Report from the NASA Advisory Council	N. Noonan		
7:00pm	Planetary Protection Program Status/Plans	J. Rummel		
7:30pm	Mars Express/Rosetta Status	Gerhard Schwehm, ESA (tent.)		
7:45pm	Mars Program Status	Orlando Figueroa, NASA HQ		
8:30pm	Mars Simulation Studies of Microbial Survival	Andrew Schuerger, DYNAMC		
9:15pm	Adjourn			
Day 2 - Esidov 20 May 2002				
	Day 2—Friday, 30 May 2003			
8:30am	Viking Planetary Protection & Current Standards	Pericles Stabekis, Windermere		
9:15am	Solar System Exploration Program Status	Colleen Hartman, NASA HQ (telephone)		
10:00am	Jupiter Icy Moons Orbiter Mission Status/Plans	Al Newhouse, NASA HQ (telephone)		
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10:30am	Break	The mouse, this is the composite of		
10:30am 10:45am	Break NRC Mars Forward Contamination Study	Pam Whitney, NRC (telephone)		
10:45am	NRC Mars Forward Contamination Study	Pam Whitney, NRC (telephone)		
10:45am 11:00am	NRC Mars Forward Contamination Study Upcoming Mars Planetary Protection Issues (Closed)	Pam Whitney, NRC (telephone)		
10:45am 11:00am 12:15pm	NRC Mars Forward Contamination Study Upcoming Mars Planetary Protection Issues (Closed) Adjourn	Pam Whitney, NRC (telephone)		
10:45am 11:00am 12:15pm 12:45pm	NRC Mars Forward Contamination Study  Upcoming Mars Planetary Protection Issues (Closed)  Adjourn  Departure to KSC Visitor's Center for KSC/CCAFS Tour	Pam Whitney, NRC (telephone)		

PPAC Meeting May 29-31, 2003
Appendix A

# PLANETARY PROTECTION ADVISORY COMMITTEE (PPAC)

Sea Oats Room, Hilton Cocoa Beach Cocoa Beach, Florida May 29–31, 2003

# **AGENDA** (continued)

Day 3—Saturday, 31 May 2003

9:00am	Committee Discussion	N. Noonan/J. Rummel
9:30am	MER Compliance Status	Laura Newlin, JPL
10:00am	MRO Planetary Protection Status	Jack Barengoltz, JPL
10:30am	Break	
10:45am	Communications Issues in Planetary Protection	Margaret Race, SETI Inst. Linda Billings, SETI Inst.
11:45am	Planetary Protection Training Course	Gary Coulter, Consultant
12:00pm	Mars Program E/PO	Michelle Viotti, JPL
12:30pm	Working Lunch	
1:00pm	Applied Molecular Methods for PP	Norman Wainwright, MBL Kasthuri Venkateswaran, JPL
2:00pm	Discussion, Applications of Molecular Methods as Standards	J. Rummel
2:30pm	Committee Discussion	
3:00pm	Adjourn	

<u>PPAC Meeting</u> <u>May 29–31, 2003</u>

Appendix B

#### PLANETARY PROTECTION ADVISORY COMMITTEE (PPAC)

Individual Members:

Dr. Norine E. Noonan, Chair Dr. John Rummel, Executive Secretary

College of Charleston NASA Headquarters

Dr. Ronald M. Atlas Dr. Colleen M. Cavanaugh

University of Louisville Harvard University

Dr. Carolyn S. Griner Dr. Debra L. Hunt Spacehab, Inc. Duke University

Dr. John F. Kerridge Mr. Alan Ladwig

Team Encounter

Dr. Debra G. B. Leonard Dr. Eugene H. Levy University of Pennsylvania Rice University

Dr. Carle M. Pieters

Brown University

Dr. Susanna Hardig Priest
Texas A&M University

Dr. George S. Robinson, III Dr. Diana Wall

Robinson & Associates, Inc.

Colorado State University

Dr. Laurie Zoloth

San Francisco State University

Representative Members:

Dr. Michael H. Carr Dr. Richard Orr

U.S. Geological Survey

U.S. Department of Agriculture

Dr. Paul Gilman Dr. Robert A. Wharton U.S. Environmental Protection Agency National Science Foundation

Dr. David Klein

NIAID, National Institutes of Health

International Representatives:

Dr. Michele Regimbald-Krnel Dr. Professor Akira Fujiwara

Space Science Program, Canadian Space Agency ISAS

Dr. Gerhard Schwehm Dr. Michel Viso

ESA/ESTEC Centre National d'Etudes Spatiales (CNES)

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Appendix C

#### PLANETARY PROTECTION ADVISORY COMMITTEE (PPAC)

Hilton Cocoa Beach Cocoa Beach, Florida May 29–31, 2003

#### MEETING ATTENDEES

#### Committee Members:

Noonan, Norine (Chair)

Atlas, Ronald

Cavanaugh, Colleen

Cavanaugh, Colleen

Cavanaugh, Colleen

Cavanaugh, Colleen

Cavanaugh

Griner, Carolyn Consultant

Kerridge, John Univ. of California, San Diego

Levy, Eugene Rice University
Pieters, Carle Brown University
Priest, Susanna Texas A&M University
Robinson, George Robinson & Associates
Rummel, John D. (Executive Secretary) NASA Headquarters
Wall, Diana Colorado State University

#### Representative Members:

Klein, David (Agency Representative)

Orr, Richard (Agency Representative)

NIH/NIAID

USDA

Regimbaud-Krnel, Michele (Internat'l Representative) Canadian Space Agency

Schwehm, Gerhard (Internat'l Representative) ESA Viso, Michel (Internat'l Representative) CNES

#### NASA Attendees:

Barengoltz, Jack JPL Campbell, James JPL

Figueroa, Orlando NASA Headquarters Hartman, Colleen (*Friday morning, by telephone*) NASA Headquarters

Kern, Roger JPL

McBride, Karen NASA Headquarters

Newlin, Laura JPL

Norris, Marian NASA Headquarters

Venkateswaran, Kasthuri JPL Viotti, Michelle JPL

#### Other Attendees

Billings, LindaSETI InstituteCoulter, GaryConsultantRace, MargaretSETI InstituteSchuerger, Andrew C.DYNAMAC Corp.Stabekis, PerryWindermere

Wainwright, Norman R. Marine Biological Laboratory

#### PLANETARY PROTECTION ADVISORY COMMITTEE (PPAC)

Hilton Cocoa Beach Cocoa Beach, Florida May 29–31, 2003

#### Presentation Slides Distributed in Hard Copy

- 1) Orlando Figueroa. *The Mars Exploration Program*, Planetary Protection Advisory Committee, May 30, 2003.
- 2) Andrew Schuerger, Rocco Mancinelli, Roger Kern, Lynn Rothschild, and Chris McKay, Survival of Endospores of Bacillus subtilis on Spacecraft Surfaces under Simulated Martian Conditions: Implications for the Forward Contamination of Mars.
- 3) P. D. Stabekis. Viking Planetary Protection & Current Standards.
- 4) Colleen N. Hartman. Solar System Exploration Division Overview to PPAC.
- 5) Alan Newhouse. Project Prometheus: Revolutionizing Solar System Exploration.
- 6) Laura Newlin et al. Mars Exploration Rove: Planetary Protection Compliance Status.
- 7) J. Barengoltz. Mars Reconnaissance Orbiter: Planetary Protection.
- 8) Margaret S. Race and Linda Billings, Communications Issues in Planetary Protection.
- 9) Gary Coulter. Planetary Protection: A Course for Practitioners
- 10) Jim Campbell. Discussion of Sample Receiving Facility Studies.
- 11) Norman L. Wainwright. Limulus Amebocyte Lysate (LAL) Test for Planetary Protection.
- 12) Kasthuri Venkateswaran. ATP as a Biomarker in Enumerating Total and Viable Microbial Burden of Spacecraft and Associated Environments.

#### Other Materials Distributed at the Meeting

- 1) Draft Request for Letter of Intent for Systems Engineering and Operational Requirements Studies for a NASA Mars Sample Receiving Facility. May 19, 2003. Jet Propulsion Laboratory.
- Study Prospectus: Preventing the Forward Contamination of Mars. Space Studies Board, National Research Council. December 2002.
- 3) Planetary Protection Class, Points of Contact. May 30, 2003.
- 4) Draft letter from John D. Rummel, NASA Office of Space Science, to editor of *Lancet* re May 24, 2003 issue containing a letter from Dr. Chandra Wickramasinghe et al.
- 5) Kasthuri Venkateswaran, Noriaki Hattori, Myuron T. La Duc, and Roger Kern. "ATP as a biomarker of viable microorganisms in clean-room facilities." *Journal of Microbiological Methods* 52: 367–377, 2003.

PPAC Meeting

# Questions from Dr. Debra Leonard in Response to the October 2002 PPAC Meeting

## **Forward contamination (higher priority)**

- 1. What conditions can extremophiles survive? What should we be concerned about?
- 2. How can extremophiles be delivered to other sites?
  - landed instruments
  - from orbiting objects
  - from crashing of orbiters.
- 3. How can bioburden be reduced and to what level (sterilization)?
- 4. How is bioburden currently being assessed for each project?
- 5. How much have we already contaminated Mars? What was the bioburden of the stuff we've already sent?
- 6. What will be the likely classification of Mars and Europa relative to NRC guidelines (the MUSES-C exercise)?
- 7. What is the current status of microbial detection, and what should NASA use to assess bioburden?

#### **Back Contamination (lower priority)**

8. Sample return handling plans require long lead times. What is being done in this regard?